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Predictors of Relapse and Overall Survival in Philadelphia Chromosome–Positive Acute Lymphoblastic Leukemia after Transplantation

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ABSTRACT

Allogeneic transplantation offers a potential cure for patients with Philadelphia chromosome–positive acute lymphoblastic leukemia (Ph+ ALL). We performed a retrospective analysis examining pretransplantation and posttransplantation prognostic factors in 90 patients with Ph+ ALL. The median age of the patients was 33 years, with slightly more than half of the patients (58%) in clinical remission at the time of transplantation. Overall, patients had a nonrelapse mortality rate of 30%, a relapse percentage of 34%, and an estimated 5-year disease-free survival rate of 30%. Pretransplantation risk factors for relapse included the expression of the p190 transcript (relative risk [RR] = 5.1; $P = .037$), evidence of morphologic disease at the time of transplantation (RR = 3.9; $P < .001$), and type of donor (RR = 2.5; $P = .015$), with patients receiving autologous or matched related transplants having the highest risk of relapse. The detection of minimal residual disease by reverse transcription polymerase chain reaction for *bcr-abl* transcripts was a significant posttransplantation risk factor for relapse (RR = 4.4; $P = .001$), with posttransplantation patients expressing the p190 transcript having the highest risk of relapse (RR = 8.7; $P = .0001$). In addition, patients with chronic extensive graft-versus-host disease showed a significantly lower risk of relapse (RR = 0.33; $P = .038$). Thus, these findings indicate that several pretransplantation and posttransplantation risk factors exist for patients with Ph+ ALL. Together, these factors can be used to improve our risk stratification of patients with Ph+ ALL who undergo transplantation, which will greatly enhance our ability to counsel these patients and potentially lead to the development of more specific treatment plans for them.

KEY WORDS

Acute lymphoblastic leukemia • Philadelphia chromosome • Transplantation • Minimal residual disease

INTRODUCTION

The Philadelphia chromosome (Ph) creates a unique fusion gene by adjoining the proximal *BCR* gene to the distal *ABL* gene [1]. Approximately 20% of adults with acute lymphoblastic leukemia (ALL) harbor the Ph chromosome and, thus, express the *bcr-abl* gene [2-4]. Although several different *bcr-abl* transcripts have been described in a variety of different leukemias, the 2 prominent types of *bcr-abl* transcripts in ALL are the p190 and p210 transcripts [5]. Patients with Ph+ ALL have a poor prognosis, with a disease-free survival (DFS) rate between 0 and 20% after standard chemotherapy [2,6,7]. For patients with a suitable donor, allogeneic transplantation offers a potential cure and a DFS between 35% and 75% [2,7-11].

Despite the encouraging results with transplantation, 30% to 50% of patients with Ph+ ALL will relapse after transplantation [8,12]. Treatments previously have been limited for patients who relapse after transplantation [13,14]; however, novel therapeutic approaches are being developed for these patients. These treatments include donor lymphocyte infusions, adoptive immunotherapy, interferon, second transplantations with nonmyeloablative regimens, and small molecular inhibitors [13-17]. These treatments may be most beneficial in patients with low tumor burden [18]. Thus, the ability to predict which patients are at high risk of relapse after transplantation and to monitor patients for the development of early relapse could be of benefit.

Multiple factors, such as conditioning regimen, donor sta-

tus, phase of disease at time of transplantation, graft-versus-host disease (GVHD), and detection of minimal residual disease (MRD) by *bcr-abl* assays, are associated with posttransplantation relapse in chronic myeloid leukemia (CML) [19-21]. Similar to CML, *bcr-abl* assays can be used to detect MRD in patients with Ph+ ALL [22]. Previously, we studied 36 patients with Ph+ ALL to examine if the detection of *bcr-abl* transcripts predicted relapse after transplantation [12]. In this study, we expand on this earlier study by examining pretransplantation and posttransplantation *bcr-abl* status as well as other possible predictors of relapse in 90 patients with Ph+ ALL who underwent hematopoietic transplantation at our Center.

MATERIALS AND METHODS

Patients

All patients with Ph+ ALL referred to the Fred Hutchinson Cancer Research Center (FHCRC) for transplantation were eligible for study. All patients were enrolled on Institutional Review Board–approved protocols with informed consent signed. Marrow and peripheral blood (PB) samples were intended to be collected before etransplantation and at posttransplantation days 21, 56, and 80 and at 6-month intervals thereafter. A total of 90 patients with Ph+ ALL underwent transplantation at FHCRC between October 1989 and January 2001. Posttransplantation samples from 36 of these patients were reported in a previous study [12].

Reverse Transcription Polymerase Chain Reaction Amplification of bcr-abl Transcripts

RNA isolation and reverse transcription polymerase chain reaction (RT/PCR) amplification were performed on each available sample as previously described [12]. The expected RT/PCR amplification products for p210 *bcr-abl* transcripts are 304 base pair (bp) or 234 bp, depending on the presence or absence of *BCR* exon b3. For the p190 *bcr-abl* transcript, the amplification product is 190 bp. Identification of specific *bcr-abl* transcript was based on the size of the RT/PCR product as determined using electrophoresis on 2% agarose. RNA samples from ALL-1 and K562 cell lines were used as positive controls for p190 and p210 *bcr-abl* transcripts, respectively. RT/PCR assays for both ALL-1 and K562 messenger RNA detected the *bcr-abl* transcripts in dilution curves as low as 10^{-5} (1 part positive control in a background of 100,000 parts normal bone marrow [BM]). RT/PCR amplification of β_2-microglobulin was used to test the quality of each patient's RNA sample. Any samples without a positive β_2-microglobulin or experiments without amplification of 10^{-5} positive controls were repeated. All experiments had a no-template reaction (negative control) to assess for contamination. A positive RT/PCR for *bcr-abl* was defined as a positive signal of the appropriate size, with appropriate negative and positive controls. Carryover and contamination were reduced through strict isolation of postamplification products and filtered pipet tips as previously described [12].

Cytogenetic Analysis

Chromosomes were prepared, and trypsin Giemsa banding patterns were analyzed from direct, 24-hour, and 48-hour unstimulated BM as previously described [12].

Direct Nucleotide Sequencing of PCR Products

Any ambiguous PCR products were verified by direct nucleotide sequencing using the 3100 Genetic Analyzer (Applied Biosystems Inc Foster City, CA) as previously described [12].

Transplantation Regimen

Most patients (n = 86; 96%) received a conditioning regimen with total body irradiation (TBI) between 1200 and 1575 cGy plus cyclophosphamide (Cy) of 120 mg/kg during 2 days. Within this group of 86 patients, 7 patients also received antithymocyte globulin and 5 patients received VP-16. For the other 4 patients, 2 patients were conditioned with BCNU, Cy, and VP-16, 1 with TBI (600 cGy), Cy, and VP-16, and 1 with a nonmyeloablative regimen of fludarabine and TBI (200 cGy). Except for the 1 patient who underwent nonmyeloablative transplantation who received GVHD prophylaxis with cyclosporine and mycophenolate mofetil, all other patients who underwent allogeneic transplantation received GVHD prophylaxis with methotrexate and cyclosporine as previously described [23].

Definitions of Outcomes and Statistics

Pretransplantation relapse was defined as any patient with ≥5% blasts in BM or abnormal blasts in PB. All other pretransplantation patients were defined as being in complete remission (CR). Posttransplantation relapse was defined as a patient with (1) ≥5% blasts in the BM, (2) leukemic blasts in the PB, (3) evidence of the Ph chromosome on cytogenetic examination in either BM or PB, or (4) extramedullary involvement of leukemia. All other posttransplantation patients were considered to be in CR. Median follow-up time was calculated as the median difference between date of last contact and transplantation date. Nonrelapse mortality (NRM) was defined as death due to anything other than relapse or the treatment of relapse. Acute GVHD (aGVHD) and chronic GVHD (cGVHD) were defined as previously described, with cGVHD being divided into limited or extensive [24,25]. Relapse rates were estimated using cumulative incidence statistics [26]. The probability of survival was estimated using the Kaplan-Meier method [27]. Survival rates were compared using the log-rank test. MRD as indicated by *bcr-abl* positivity was used as a time-dependent covariate in a Cox regression model comparing the hazard rates of relapse [28]. Therefore, if a patient had several posttransplantation RT/PCR measurements for *bcr-abl*, the patient's *bcr-abl* status might change over time in the analysis. In addition, the time to relapse data were treated as left-truncated, so that a patient did not enter the risk set until the first RT/PCR was performed. Patients who were positive for *bcr-abl* on a sample also showing morphological or cytogenetic relapse were classified according to the *bcr-abl* status on the previous RT/PCR. We report *P* values from the Wald test for effects in the Cox regression models.

RESULTS

Patients and Clinical Outcomes

A total of 90 patients with Ph+ ALL underwent transplantation at FHCRC between October 1989 and January 2001 (Table 1). The median age of the patients was 33 years (range, 2-56 years), with a median time of follow-up for all 90 patients of 280 days (range, 4-4004 days). The median follow-up for surviving patients was 1516 days (range, 29-4004 days). More

Table 1. *Patient Characteristics and Type of Transplantation*

	Patients
No. of patients	**90**
Sex (No., percent male)	**52 (58)**
Age (median, range in years)	**33 (2-56)**
Phase of disease (No., percent)	
Remission	**52 (58)**
Relapse	**38 (42)**
Donor match (No., percent)	
Unrelated	**37 (41)**
Related-matched	**31 (34)**
Related-mismatched	**14 (16)**
Autologous or syngeneic	**8 (9)**
Conditioning (No., percent)	
≥1200 cGY	**86 (96)**
Chemotherapy only or <1200 cGY	**4 (4)**

than half of the patients (N = 52; 58%) were in CR at the time of transplantation. Twenty-seven of 90 (30%) patients died without relapse, and 31 of 90 (34%) patients relapsed. The DFS and overall survival (OS) rates for the 90 patients were similar, with both approximately 30% at 5 years (Figure 1).

Pretransplantation Predictors of Relapse

Patients in first CR or subsequent CR had a better clinical outcome than those patients in relapse at the time of transplantation (Table 2). In univariate analyses of the 90 patients, donor type, phase of disease, and year of transplantation had a significant impact on the relative risk (RR) of relapse (Table 3). In multivariate analyses, clinical relapse at the time of transplantation was the strongest predictor of relapse after transplantation (RR = 3.9; $P < .001$). Donor type (auto/twin/matched related donor versus mismatched related donor/unrelated) remained predictive (RR = 2.9; $P = .005$) after controlling for remission status and year of transplantation. The increased risk of relapse in patients who have undergone transplantation recently was probably because of a higher percentage of patients undergoing transplantation in clinical relapse during this time period (1996 to 2001 = 68% versus 1989 to 1995 = 49%; Chi-squared test, $P = .065$). Although age did not have an impact on relapse (Table 3), the age of the patient appeared to influence survival. In a multiple Cox regression model accounting for remission status, patients aged 33 years or older had a higher risk of death compared with younger patients (RR = 1.7; $P = .042$).

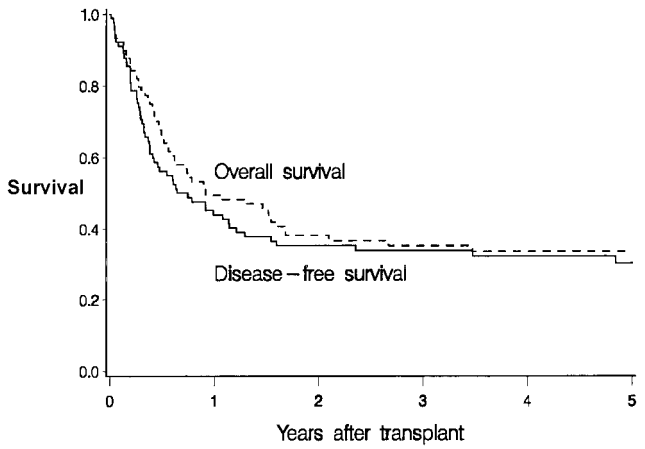

Figure 1. OS and DFS estimates for all patients (n = 90). The graph shows that OS and DFS curves essentially overlap one another, indicating that most patients who relapse die. In addition, the curves begin to plateau after 2 years, suggesting that long-term survival is possible.

Table 2. *Outcomes for 90 Patients With Ph+ ALL*

	Auto/ Twin	MRD	MMRD	URD	Total (%)
First CR (n = 42)					
Alive without relapse	**2**	**6**	**2**	**13**	**23 (55)**
Relapse	**2**	**5**	**0**	**1**	**8 (19)**
NRM	**0**	**4**	**2**	**5**	**11 (26)**
Total	**4**	**15**	**4**	**19**	**42 (100)**
Subsequent CR (n = 10)					
Alive without relapse	**1**	**0**	**0**	**3**	**4 (40)**
Relapse	**0**	**1**	**0**	**2**	**3 (30)**
NRM	**0**	**1**	**0**	**2**	**3 (30)**
Total	**1**	**2**	**0**	**7**	**10 (100)**
Relapse (n = 38)					
Alive without relapse	**0**	**1**	**2**	**2**	**5 (13)**
Relapse	**3**	**9**	**4**	**4**	**20 (53)**
NRM	**0**	**4**	**4**	**5**	**13 (34)**
Total	**3**	**14**	**10**	**11**	**38 (100)**
All patients (n = 90)					
Alive without relapse	**3**	**7**	**4**	**18**	**32 (36)**
Relapse	**5**	**15**	**4**	**7**	**31 (34)**
NRM	**0**	**9**	**6**	**12**	**27 (30)**
Total	**8**	**31**	**14**	**37**	**90 (100)**

MRD indicates matched related donor; MMRD, mismatched related donor; URD, unrelated donor; Auto, autologous transplant.

Pretransplantation *bcr-abl* Expression as a Predictor of Relapse

We investigated whether *bcr-abl* status before transplantation was predictive of relapse. Of the 90 patients, 51 patients had

Table 3. *Univariate Analyses of Pretransplantation Predictors of Relapse*

	n	Relapse n (%)	RR	95% CI	P*
Age					
<33 y	**45**	**15 (33)**	**1.0**		
≥33 y	**45**	**16 (36)**	**1.4**	**0.70-2.9**	**.33**
Donor status					
Unrelated	**37**	**7 (19)**	**1.0**		
Mismatched related	**14**	**4 (29)**	**1.7**	**0.49-5.8**	**.40**
Matched related	**31**	**15 (48)**	**3.0**	**1.2-7.5**	**.015**
Auto or syngeneic	**8**	**5 (63)**	**3.1**	**0.98-9.7**	**.055**
Transplantation year					
1989-1995	**49**	**12 (24)**	**1.0**		
1996-2001	**41**	**19 (46)**	**2.3**	**1.1-4.7**	**.032**
Phase of disease					
Clinical remission	**52**	**11 (21)**	**1.0**		
Clinical relapse	**38**	**20 (53)**	**4.0**	**1.9-8.4**	**.0003**
MRD					
bcr-abl negative	**17**	**2 (12)**	**1.0**		
bcr-abl positive	**34**	**15 (44)**	**4.0**	**0.92-18**	**.065**

CI indicates confidence interval.

**P* calculated using Wald test in Cox regression model.

a RT/PCR test for *bcr-abl* expression immediately before transplantation, with the remaining 39 patients refusing the test or with the test not being ordered on their pretransplantation evaluation. These 51 patients were not statistically different with respect to age, gender, donor type, disease phase, conditioning regimen, or outcomes compared with the 39 patients without a pretransplantation *bcr-abl* assay. The majority of the 51 patients (n = 34; 67%) had a positive *bcr-abl* assay immediately before transplantation, whereas the other 17 patients showed no evidence of *bcr-abl* expression using RT/PCR assays. Half (n = 17) of the 34 patients with positive *bcr-abl* assays were classified as being in clinical remission based on morphologic examination of BM immediately before transplantation. The incidence of relapse for the 17 patients who were *bcr-abl*–negative before transplantation was 12%, compared with 44% in the 34 patients with a positive pretransplantation *bcr-abl* assay. After adjusting for clinical stage of disease at transplantation, pretransplantation *bcr-abl*–positivity conferred a marginally significant increased RR of relapse (RR = 3.5; $P = .095$), suggesting that MRD as indicated by a positive *bcr-abl* test may be an independent pretransplantation predictor of relapse. Patients in CR without MRD (as determined using *bcr-abl* assay) had the highest DFS, followed by patients in CR with MRD, and then patients in known clinical relapse (Figure 2).

Posttransplantation Predictors of Relapse

We also examined posttransplantation factors that might influence the RR of relapse and OS in patients with Ph+ ALL. aGVHD and cGVHD were treated as time-dependent covariates, with cGVHD analyzed in patients who survived to 100 days after transplantation. In univariate analyses, aGVHD (RR = 0.67; $P = .44$) was not significantly associated with ALL relapse, whereas extensive cGVHD (RR = 0.44; $P = .093$) was associated with a marginally significant reduction in relapse (Table 4). After adjusting for pretransplantation risk factors (donor type, phase of disease at transplantation, and transplantation year), patients with chronic extensive GVHD showed a statistically significantly lower risk of relapse (RR = 0.33; $P = .038$).

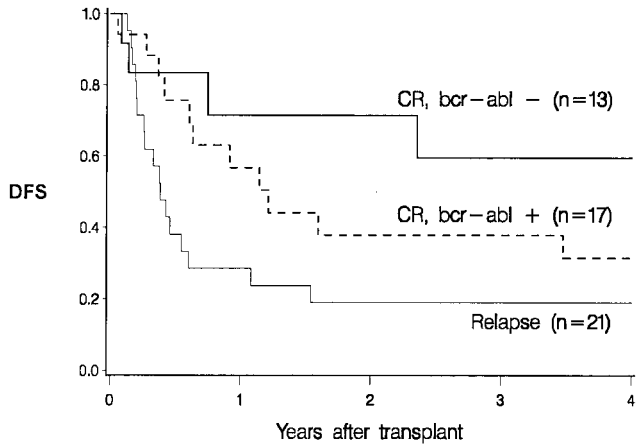

Figure 2. DFS by pretransplantation RT/PCR and disease status. Kaplan-Meier estimates are shown for 3 patient groups: CR and no evidence of *bcr-abl* expression (CR, *bcr-abl* −), CR and evidence of *bcr-abl* expression (CR, *bcr-abl* +), and relapse. The difference in DFS between patients in CR and relapse was statistically significant ($P = .017$). The differences between the relapse group and each of the CR groups by *bcr-abl* expression were not statistically significant, but the groups are shown for descriptive purposes.

Table 4. *Univariate Analyses of Posttransplantation Predictors of Relapse*

	n	Relapse n (%)	RR	95% CI	P*
aGVHD					
Grade 0-I	**18**	**5 (28)**	**1.0**		
Grade II-IV	**54**	**20 (37)**	**0.67**	**0.25-1.8**	**.44**
cGVHD					
None or limited	**31**	**17 (55)**	**1.0**		
Extensive	**34**	**7 (21)**	**0.45**	**0.18-1.1**	**.093**
MRD					
bcr-abl negative	**31**	**7 (23)**	**1.0**		
bcr-abl positive	**33**	**15 (45)**	**4.4**	**1.8-11**	**.0013**

**P* calculated using Wald test in Cox regression model.

Posttransplantation *bcr-abl* Expression as a Predictor of Relapse

Sixty-four patients had at least 1 *bcr-abl* assay after transplantation and before clinical relapse. Compared with the 26 patients who did not have *bcr-abl* data after transplantation, these 64 patients were not significantly different with respect to gender, age, donor type, or conditioning regimen. Patients without *bcr-abl* data after transplantation had a significantly higher incidence of NRM compared with those patients with a posttransplantation *bcr-abl* test (62% versus 17%, Fisher exact test; $P < .001$).

Thirty-three of the 64 patients (52%) had positive *bcr-abl* assay at some point after transplantation and before clinical relapse. Of the 33 patients with a *bcr-abl* positive test after transplantation, 15 patients (45%) relapsed and 6 died without relapse. The median time from the first positive *bcr-abl* assay to relapse was 75 days. Twelve patients with a positive *bcr-abl* assay after transplantation remained alive without clinical disease, with a median follow-up of 2453 days (range, 714-4004 days). Nine of the 12 patients had only 1 or 2 positive assays and thereafter had negative test results up through the date of last contact. Two patients had no more *bcr-abl* assays after their last positive test. Only 1 patient remained positive from day 21 to day 1042 (day of last test). The 12 patients who were positive for *bcr-abl* and did not relapse were somewhat more likely to have had cGVHD compared with the other 45 patients with posttransplantation *bcr-abl* data who survived to day 100 (75% versus 47%; $P = .11$). In the 31 patients who were persistently negative for *bcr-abl*, 7 patients (23%) relapsed and 5 died without relapse. The median time from the last negative *bcr-abl* assay to relapse was 146 days, and detection of *bcr-abl* was a significant posttransplantation predictor of clinical relapse (RR = 4.4; $P = .001$). After adjusting for donor type and disease phase at the time of transplantation, the presence of MRD posttransplantation continued to be significantly associated with relapse (RR = 3.0; $P = .029$). The 5-year DFS rate for patients with positive RT/PCR within 100 days posttransplantation was statistically lower than those without a positive RT/CR (29% versus 57%; $P = .007$) (Figure 3).

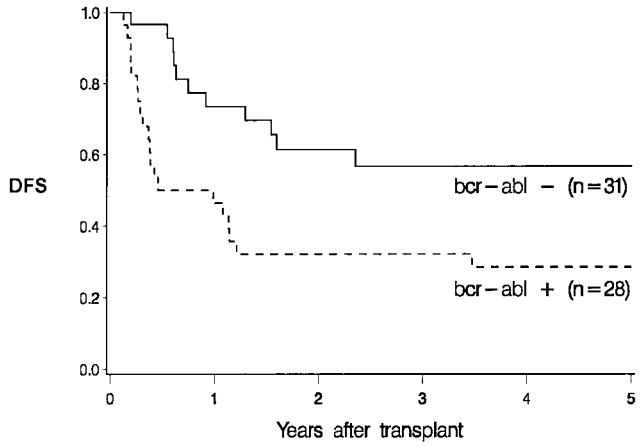

Figure 3. DFS by RT/PCR from 100 days posttransplantation, stratified by evidence of *bcr-abl* expression. Risk stratification by expression of *bcr-abl* within the first 100 days posttransplantation shows a relationship between MRD and DFS ($P = .007$). Kaplan-Meier estimates are shown for 2 patient groups: no evidence of *bcr-abl* expression (*bcr-abl* −) and evidence of *bcr-abl* expression (*bcr-abl* +) within the first 100 days posttransplantation.

Type of *bcr-abl* Transcript as a Predictor of Relapse

The type of *bcr-abl* transcript detected before transplantation and after transplantation influenced the risk of relapse. Of the 51 patients with a pretransplantation *bcr-abl* assay, patients expressing the p190 *bcr-abl* transcript had a significantly increased risk of relapse (RR = 5.1; $P = .037$) compared with patients without evidence of *bcr-abl* before transplantation (Table 5). Although the p210 *bcr-abl* transcript did confer a higher risk of relapse, this risk of relapse was not statistically significant (Table 5). Likewise, expression of the p190 *bcr-abl* transcript posttransplantation was associated with a statistically increased risk of relapse posttransplantation (RR = 8.7; $P < .001$) compared with those without evidence of the *bcr-abl* expression (Table 5). Again, the magnitude of the RR for the p210 *bcr-abl* expression was not as great and did not reach statistical significance (Table 5).

DISCUSSION

We investigated possible pretransplantation and posttransplantation risk factors for relapse in 90 patients with Ph+ ALL who underwent transplantation at a single institution between 1989 and 2001. Overall, the NRM incidence was 30%, and 34% of patients relapsed. The overall DFS rate at 5 years was 30% (Figure 1). Pretransplantation risk factors for relapse included the following: (1) type of donor (patients receiving an autologous and/or matched related donor had the highest risk of relapse); (2) evidence of clinical disease at the time of transplantation; and (3) expression of *bcr-abl*, particularly the p190 transcript. The best pretransplantation prognosis was found in patients without evidence of morphological or molecular disease. Patients with evidence of molecular disease but without morphological disease had an intermediate prognosis, whereas those patients with evidence of morphological disease had the worst prognosis (Figure 2). Evidence of MRD, especially with the expression of the p190 transcript, and an absence of cGVHD were significant posttransplantation risk factors for relapse.

In our study, 38 allogeneic patients underwent transplantation in first CR (Table 2), with a median follow-up of 482 days (range, 13-3822 days). Twenty-one of these patients (58%) remained alive without relapse. Our results in patients with Ph+ ALL who underwent transplantation in first CR are similar to previously reported studies. For example, Synder et al. [11] examined 23 patients with Ph + ALL (median age, 25 years) who underwent transplantation in first CR with matched related donors. Synder et al. reported that 14 of the 23 patients (61%) were alive without disease a median of 34 months after transplantation. We were able to further risk stratify our patients in CR by evidence of MRD. Those patients in CR without evidence of pretransplantation MRD had the lowest incidence of relapse (12%) and the highest DFS rate (Figure 2). Transplantation studies in ALL patients without a Ph chromosomal abnormality also have found that pretransplantation MRD status influences relapse risk [29-31]. These results raise the question of whether pretransplantation patients with MRD should receive additional treatment before transplantation.

The transplantation results for patients with Ph+ ALL in CR are in stark contrast to the transplantation results for patients with evidence of morphological disease at the time of transplantation. In our study, most patients who were not in CR at the time of transplantation either relapsed (17 of 35, 49%) or died of other causes (13 of 35, 37%). Only 5 of 35 patients (14%) remained alive without evidence of disease. Together, the results from this study add to the growing body of evidence that disease burden at the time of transplantation is a significant predictor of relapse after transplantation in patients with ALL and that transplantation in first CR appears to be the preferred treatment option for patients with Ph+ ALL.

Previous studies have shown that the detection of MRD after transplantation is highly predictive of relapse [12,19,32]. This study confirms our previous results, reiterating the importance of MRD monitoring after transplantation. In our study, 12 of the 33 (36%) patients who had expression of the *bcr-abl* transcript after transplantation remained free of clinical disease at the time of this report. Nine of these 12 patients had only 1 or 2 positive tests and then became negative for the *bcr-abl* transcript up through the date of last contact. Two patients had no additional tests. Only 1 patient had evidence of long-term, persistent *bcr-abl* transcript at the time of last contact. The 12

Table 5. *Relapse Risk by bcr-abl Type*

	n	Relapse, n (%)	RR	95% CI	P*
Before transplantation					
None	17	2 (12)	1.0		
p190	19	9 (47)	5.1	1.1-24	.037
p210	8	3 (38)	2.9	0.48-17	.25
p190 and p210	7	3 (43)	3.3	0.55-20	.19
After transplantation					
None	31	7 (23)	1.0		
p190	14	7 (50)	8.7	3.0-25	.0001
p210	12	4 (33)	2.2	0.65-7.7	.20
p190 and p210	7	4 (57)	5.0	1.5-17	.010

*P calculated using Wald test in Cox regression model.

patients expressing *bcr-abl* transcripts without relapse were more likely to have had cGVHD compared with the other patients with posttransplantation *bcr-abl* data (75% versus 47%; $P = .11$), suggesting that the immune system may be playing a role in preventing relapse in patients with Ph+ ALL with MRD. Indeed, cGVHD was associated with a lower risk of relapse in multivariate analyses (RR = 0.44; $P = .038$). Quantitative RT/PCR, which can provide a determination of the level of *bcr-abl* expression, may be a more accurate method than qualitative RT/PCR to assess the burden of disease and risk of relapse. Studies have already shown that the posttransplantation burden of disease, as measured by quantitative RT/PCR for *bcr-abl*, is highly predictive of relapse in patients with CML after transplantation [33].

Seven of 31 patients (23%) relapsed without having a previously positive RT/PCR for *bcr-abl*. Although attempts were made to screen these patients at days 21, 56, and 80 and at 6-month intervals, some patients missed blood draws because of various reasons. Patients with negative *bcr-abl* tests relapsed a median of 146 days after their last negative test, whereas those patients with a positive *bcr-abl* assay relapsed a median of 75 days after their first positive test. This suggests that we may have been able to identify additional patients before clinical relapse if we had screened them at more frequent intervals.

Previously, we found that posttransplantation expression of the p190 transcript carried a higher RR of relapse than expression of the p210 [12]. Recently, Kroger et al. [34] were unable to demonstrate significant clinical differences when stratifying 19 patients according to type of transcript expressed before transplantation. In the current analysis, we found that expression of p190 in both the pretransplantation and posttransplantation setting was associated with a statistically significantly higher risk of relapse than no expression, whereas the expression of p210 did not confer a significantly higher risk. Small sample size could account for some of the clinical differences between p190 and p210, but several lines of research predict that clinical outcomes might differ depending on the type of transcript expressed. In vitro assays, including experiments in immature lymphoid cells, have shown that the p190 protein has an increased tyrosine kinase activity compared with the p210 protein [35,36]. In murine models, the p190 also was found to cause a more aggressive form of leukemia [37,38]. Thus, the biology of the 2 transcripts would suggest that p190 would confer a higher rate of therapy resistance and a worse prognosis. Our findings, combined with the biology data, indicate that differential treatment strategies may be necessary for patients expressing p190 versus p210.

In summary, multiple pretransplantation and posttransplantation risk factors are available to help risk stratify patients with Ph+ ALL undergoing transplantation. The burden of disease at the time of transplantation and detection of MRD after transplantation are predictive of relapse [29-31]. These data suggest a future where patients are risk stratified before transplantation, and the intensity of conditioning and immunosuppression are tailored to the patient based on pretransplantation risk factors. "Low-risk" patients (no evidence of MRD) may be eligible for less intense conditioning regimens, such as nonmyeloablative transplantation, whereas "high-risk" patients (MRD or overt clinical disease) would be eligible for more aggressive immunosuppression tapers or even posttransplantation consolidation with novel treatment approaches, including small molecular inhibitors aimed at blocking *bcr-abl* function, such as STI-571 (Gleevec, Novartis, East Hanover, NJ). In addition, our results reiterate the need to better define predictors of relapse and to develop targeted treatment approaches. However, many of these posttransplantation interventions probably will require a more precise understanding of the biological (eg, p190 versus 210) mechanisms that determine relapse.

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